

WHAT IS CLAIMED IS:

1. An isolated polynucleotide encoding an enzyme having astaxanthin synthase activity which catalyzes the reaction of beta-carotene to astaxanthin.

2. An isolated polynucleotide according to claim 1 wherein the reaction is catalyzed in *P. rhodozyma*.

3. An isolated polynucleotide according to claim 1 which is selected from

(a) a nucleotide sequence which encodes an enzyme having the amino acid sequence shown in SEQ ID NO: 1, or

(b) a nucleotide sequence which encodes (i) an allelic variant of SEQ ID NO:1 or (ii) an enzyme having the polypeptide sequence of SEQ ID NO:1 with one or more amino acid insertions, deletions, and/or substitutions, which variant has astaxanthin synthase activity.

3. An isolated polynucleotide according to claim 1 which is selected from the group consisting of:

(i) SEQ ID NO: 2;

(ii) a nucleotide sequence which, because of the degeneracy of the genetic code, encodes an astaxanthin synthase having the same amino acid sequence as that encoded by SEQ ID NO:2; and

(iii) a nucleotide sequence which hybridizes to the complement of the nucleotide sequence from i) or ii) under standard hybridizing conditions (50% v/v formamide, 5X SSC, 2% w/v blocking agent, 0.1% N-lauroylsarcosine, 0.3% SDS at 42EC overnight).

4. An isolated polynucleotide according to claim 1 which is selected from the group consisting of:

(i) SEQ ID NO: 3;

(ii) a nucleotide sequence which, because of the degeneracy of the genetic code, encodes an astaxanthin synthase having the same amino acid sequence as that encoded by SEQ ID NO:3; and

(iii) a nucleotide sequence which hybridizes to the complement of the nucleotide sequence from i) or ii) under standard hybridizing conditions (50% v/v formamide, 5X SSC, 2% w/v blocking agent, 0.1% N-lauroylsarcosine, 0.3% SDS at 42EC overnight).

→ 5 6 5. A vector or plasmid comprising a polynucleotide which encodes an enzyme having astaxanthin synthase activity.

7 6. A vector or plasmid according to claim 5 wherein the polynucleotide encodes SEQ ID NO:1.

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8 7. A vector or plasmid according to claim 5 wherein the polynucleotide is SEQ ID NO:2.

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9 8. A vector or plasmid according to claim 5 wherein the polynucleotide is SEQ ID NO:3.

10 9. A host cell transformed or transfected with a polynucleotide which encodes an enzyme having astaxanthin synthase activity.

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11 10. A host cell according to claim 9 wherein the polynucleotide encodes a polypeptide having the sequence of SEQ ID NO:1.

12 11. A host cell according to claim 9 wherein the polynucleotide is SEQ ID NO:2.

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13 12. A host cell according to claim 9 wherein the polynucleotide is SEQ ID NO:3.

14 13. A host cell according to claim 9 which is transfected or transformed with a vector or a plasmid comprising: (a) a polynucleotide which encodes the polypeptide of SEQ ID NO:1; (b) SEQ ID NO:2; and/or (c) SEQ ID NO:3.

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15 14. A recombinantly-produced polypeptide encoded by a polynucleotide which is SEQ

ID NO:2 or SEQ ID NO:3.

16 15. A process for producing a polypeptide having astaxanthin synthase activity comprising culturing a host cell transformed with a polynucleotide which encodes an enzyme having astaxanthin synthase activity under conditions conducive to produce the enzyme.

17 16. A process according to claim 15 wherein the polynucleotide encodes the polypeptide of SEQ ID NO:1.

18 17. A process according to claim 15 wherein the polynucleotide is SEQ ID NO:2.

19 18. A process according to claim 15 wherein the polynucleotide is SEQ ID NO:3.

20 19. A process for producing astaxanthin comprising introducing one or more isolated polynucleotides encoding an enzyme having astaxanthin synthase activity into an appropriate host organism; cultivating the organism under conditions conducive to produce astaxanthin; and recovering astaxanthin from the culture.

21 20. A process according to claim 19 wherein the polynucleotide is selected from the group consisting of a polynucleotide encoding SEQ ID NO:1, SEQ ID NO:2, and SEQ ID NO:3.

22 21. A process for producing astaxanthin comprising contacting beta-carotene with a polypeptide having astaxanthin synthase activity in the presence of an electron donor in a reaction mixture containing a reconstituted membrane.

23 22. A process according to claim 21, wherein the polypeptide is present in the form of a reconstituted membrane which is prepared from a biological membrane.

24 23. A process according to claim 22 wherein the membrane is a microsome or a

mitochondrial membrane.

25 24. A process according to claim 21 wherein the polypeptide is present in the form of a reconstituted artificial membrane.

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26 25. A process according to claim 24 wherein the reconstituted artificial membrane is a liposome.

27 26. A process according to claim 21, wherein the electron donor reduces a reaction  
10 center of the astaxanthin synthase.

28 27. A process according to claim 26 wherein the electron donor is cytochrome P450 reductase

15 29 28. An isolated polynucleotide encoding a polypeptide which is SEQ ID NO:1.

30 29. An isolated polynucleotide consisting of SEQ ID NO:2.

31 30. An isolated polynucleotide consisting of SEQ ID NO:3.

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32 31. A recombinantly-produced polypeptide having astaxanthin synthase activity which is SEQ ID NO:1.

33 32. A vector comprising a polynucleotide selected from the group consisting of a  
25 polynucleotide which encodes SEQ ID NO:1, SEQ ID NO:2, and SEQ ID NO:3.

34 33. A host cell transformed with the vector of claim 32.

35 34. A process for producing astaxanthin comprising: (a) cultivating in a suitable  
30 culture medium a recombinantly produced host cell containing a polynucleotide which encodes a polypeptide having astaxanthin synthase activity.

35. A process according to claim 34 wherein the polynucleotide encodes a polypeptide which is SEQ ID NO:1.

36. A process according to claim 34 wherein the polynucleotide is SEQ ID NO:2.

37. A process according to claim 34 wherein the polynucleotide is SEQ ID NO:3.

38. A process according to claim 34 further comprising isolating the astaxanthin synthase from the host cell or the culture medium.

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